…ranaia (FA) protein dysfunction and cytogenetic instability …ident prostate cancer cells. Susan A. Speckhart, Winifred …ari, Susan B. Olson, and Grover C. BagbyJr. *OHSU Cancer …*

…n exists between tumor aggressiveness and cytogenetic insta… …osomal defects represent an unfavorable prognostic factor in …cluding cancer of the prostate. In this tumor type, accumu… …rmities has correlated positively with outgrowth of androgen… …While some molecular causes of genetic instability in cancers of …hematopoietic tissues are recognized, the mechanisms that …bility in prostate cancer cells are unknown. For decades, a …drome Fanconi anemia has been recognized to predispose …marrow failure, leukemia, and solid tumors. There are eight …groups from which 7 genes have been cloned and character… …ducts collaborate with each other and with BRCA1 and …ther detect and/or repair DNA cross links and double strand …proteins (FANCA, C, E, F, and G) form a nuclear complex …no-ubiquitinylation of the sixth (FANCD2). This post-trans… …ired for FANCD2 to associate with BRCA1 in DNA damage …venth FA gene is BRCA2 of which C-terminal truncating …es. Testing the hypothesis that abnormalities of the FA path… …ability in prostate cancer cells, we quantified the function of …ee prostate cancer cell lines: DU145 and PC3 (both androgen …ogenetically complex) and LNCaP (androgen dependent). …trophoresis, and immunoblotting with polyclonal antisera to …2 we determined that two cell lines demonstrated abnormal… …ay. In the DU145 cell line, FANCD2-S (not monoubiquiti… …it FANCD2-L (ubiquitinated) was not implicating dysfunc… …FANCA, C, E, F, or G. In the PC3 cell line, BRCA2, …CD2-L were detectable but significantly decreased. Dysfunc… …ty in both of these cell lines was confirmed in the mitomycin …kage test. The LNCaP cell line was mitomycin resistant by …nd BRCA2 and FANCD2S and L were normal. We suggest …e FA pathway may represent an important progression factor …id genetic instability in prostate cancer cells.

…of transcription factor Oct-1 by Ku/DNA-dependent pro… …nse to DNA damage. Caroline Schild-Poulter, Amy Shih, …n, Jeff C. Kochan, Sebastien Soubeyrand, and Robert JG …*Health Research Institute, Ottawa, Ontario, Canada.*

…ers a signal transduction cascade leading to cell cycle arrest and …A repair pathways. The Ku/DNA-dependent protein kinase …is a necessary component of the non-homologous end-joining …pathway, through its direct participation in the DNA repair …s also been suggested to be involved in the signaling cascade, …a function is still lacking. We have recently documented an …-u and transcription factor Oct-1, an ubiquitous homeodo… …or. Oct-1 has been implicated in the regulation of transcrip… …cluding histone H2B and the immunoglobulin genes, and …egulate viral replication. Oct-1 is known to be differentially …the cell cycle but the status of its phosphorylation after …tly unknown. Here we show that Oct-1 is up-regulated in …n one hour following treatment by various dose of ionizing …hway dependent on Ku and DNA-PK. This up-regulation …l lines lacking either Ku or DNA-PK and was dependent on …l region comprising its trans-activation domain and on the …h which Oct-1 interacts with Ku70. Intriguingly, exposure to …hibited the ability of Oct-1 to activate transcription from …l templates. Moreover, endogenous H2B mRNA was found …following DNA damage in a Ku and DNA-PK-dependent …in vitro phosphorylation of full-length Oct-1 revealed that …ates several residues in the glutamine-rich N-terminal region …ng to the transactivation domain. These results suggest that …plex modulate Oct-1 function following DNA damage. The …itution of the Oct-1 residues targeted by DNA-PK are cur… …ed.

…ity of mtDNA regions to DNA damage and mutation. …th Mambo, Xiangqun Gao, Paul Talaly, and David Sidran… …*University SOM, Baltimore, MD.*

…ations occur in a wide variety of degenerative diseases and …ought to be more susceptible to DNA damage and conse… …rate of mutation than nuclear DNA. Many of the somatic mtDNA mutations in human cancers are located in the D-loop and in particular in a poly cytidine stretch (C-tract) termed D310. The D310 region exhibits polymorphic length variation among individuals and has been described to be a "hot spot" for somatic mutations in many cancer types. We analyzed mtDNA integrity and damage repair after exposure to a DNA UV-mimetic damaging agent using real time quantitative PCR. The mtDNA integrity of different regions was analyzed by calculating the mtDNA to nuclear DNA (mtDNA/nDNA) ratio. A lower mtDNA/nDNA ratio represents less initial template denoting a decrease in the integrity of mtDNA. The results showed that mtDNA damage profile was region dependant. The tRNAG was the least affected region whereas the D-loop was the most sensitive to damage. Furthermore, D310 was the most sensitive region to damage compared to the other D-loop regions. When we examined the temporal repair of mtDNA following exposure to DNA damaging agents we found that the D-loop and in particular D310 harbored delayed repair when compared with the Cox II and tRNAG regions. Delayed repair gave rise to common D310 C-tract frameshift mutations. Our results demonstrate that the D-loop (and in particular the D310 region) is highly susceptible to mutations due to its vulnerability to DNA damage coupled with inefficient repair of this region. The susceptibility of the D310 to mutations may also explain the high frequency of D310 somatic mutations detected in many tumor types.

#1197 Reciprocal translocations in mucoepidermoid carcinoma. Giovanni Tonon, Kristen Stover, Raluca Yonescu, Anna Roschke, Frederic J. Kaye, and Ilan R. Kirsch. *Genetics Branch, National Cancer Institute, Bethesda, MD.*

Mucoepidermoid carcinoma is the most common human malignant salivary gland tumor and can arise from both major salivary glands and minor salivary glands including sites within the pulmonary tracheobronchial tree. We performed Comparative Genomic Hybridization (CGH) and Spectral Karyotyping (SKY) on two tumor cell lines that were generated from tumors located in either the parotid gland or the lungs. In both cell lines, CGH showed a gain at the chromosomal band 7p21 and SKY demonstrated the presence of the previously reported balanced translocation t(11;19)(q21;p12). Multiple chromosomal rearrangements were also present in both cell lines, including four reciprocal translocations in cell line H292 (t(1;16), t(6;8)x2 and t(11;19)), and four reciprocal translocations in cell line H3118 (t(1;7), t(3;15), t(7;15), t(11;19)). A review of the literature confirmed the presence of a pattern predominated by reciprocal translocations in other reported cases of mucoepidermoid carcinomas analyzed with standard G-banding techniques, as well as distinct benign salivary gland tumors like pleomorphic adenomas and Whartin tumor. Among solid tumors, only some pediatric sarcomas present a similar cytogenetic feature with frequent reciprocal chromosomal rearrangements. In addition to the shared characteristic t(11;19), fluorescence in situ hybridization with BAC clones showed that one partner of two distinct translocations in the two cell lines presented a breakpoint in an interval of less than two megabases, at 5p15. The involvement of similar chromosomal bands in breakpoints in these two cell lines suggests that this additional region may be selected or predisposed to chromosomal rearrangements in this tumor type. The presence of multiple reciprocal translocations in selected benign and malignant salivary gland tumors may also suggest a mechanism within mucous/serous glands mediating chromosomal rearrangements

#1198 Increased gene copy number may alter E2F1 activity in melanoma growth. Mark A. Nelson, Anthony Beas, Anne-Christine Goulet, David T. Lowry, Steven H. Reynolds, Amy M. Jefferson, Jamie R. Senft, and Linda Sargent. *Department of Pathology, The University of Arizona, Tucson, AZ and Centers for Disease Control, National Institute for Occupational Safety and Health, Morgantown, WV.*

The identification of recurring translocations and unique chromosome break points in melanoma will aid in the identification of the genes that are important in the neoplastic process. Previous studies by our group identified translocations der(12)t(12;20) in malignant melanoma cells. The transcription factor E2F1 maps to 20q11. Deregulated E2F transcriptional activity has been associated with the autonomous growth of melanoma cells, but the molecular basis has not yet been elucidated. To this end, we investigated E2F1 gene copy number and structure in nine different early passage human melanoma cell lines. Fluorescent in situ hybridization analysis using a specific E2F1 probe indicated increased E2F1 gene copies in several melanoma cell lines compared to normal melanocytes. The FISH observations were confirmed by comparative genomic hybridization array to BAC clones and Southern Blot analysis. In addition, Western blot analysis demonstrated increased E2F1 and DP-1 protein levels in 8 out of 9 melanoma cells compared to normal melanocytes. These data suggests that the release of E2F activity by elevated E2F1 gene copy numbers may play a functional role in melanoma growth. (Supported by CA 70145).

The World's Premier Multidisciplinary Meeting.

Presenting Cutting-Edge Laboratory, Translational, and Clinical Cancer Research.

AACR *American Association for Cancer Research*

AB-03-CC4/6 ✓

94th Annual Meeting 2003

April 5-9, 2003 • Metro Toronto Convention Centre
Toronto, Ontario, Canada

Proceedings

Volume 44
March 2003